A NOVEL FOLD AND A NEW PROTEIN FAMILY – STRUCTURAL CHARATERIZATION OF YER067W PROTEIN OF S. *cerevisiae*

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We have undertaken a small scale structural genome project focusing on unknown ORFs up regulated by high hydrostatic pressure stress in *S. cerevisiae*. Among the 9 proteins expressed in *E. coli*, Yer067w presented the best 1D ^{1}H-Nuclear Magnetic Ressonance (NMR) spectrum, with sharp lines and large chemical shift dispersion, characteristic of a well-structured protein. However, this protein was refractory to the crystallization conditions tested. As the 10 first amino acids of Yer067w are mainly polar charged residues that probably form a flexible moiety, we removed those residues by cloning an N-shortened $_{11V-161N}$Yer067w. This version conserved the overall fold, as confirmed by NMR, but the enhanced stability contributed to a successfully crystallization. The structure of SeMet-labeled $_{11V-161N}$Yer067w, solved by Multiple Anomalous Diffraction at 1.7 Å resolution, revealed an alpha-beta fold that could not be classified in any of the families annotated in the Structural Classification Of Proteins database. A structure based search using Secondary Structure Matching server, retrieved only 2 proteins with more than 60% match, both of unknown function. A primary sequence search showed that Yer067w belongs to well conserved family exclusive of Ascomycetes. Furthermore, the alignment of these 11 unknown relatives revealed that the 100% conserved amino acids are mainly clustered in a cleft. However, this putative biding site could not be identified in another structural model deposited in the Protein Data Bank. As the yeast deleted for Yer067w presented important phenotypes, the functional characterization of this protein will bring new insights, both for structural biology and for the comprehension of cell metabolism. Support: CNPq